BEID Workshop

March 29, 2006

Development of ABthrax, a fully human monoclonal antibody directed against Bacillus anthracis protective antigen (PA), for the treatment of inhalational anthrax

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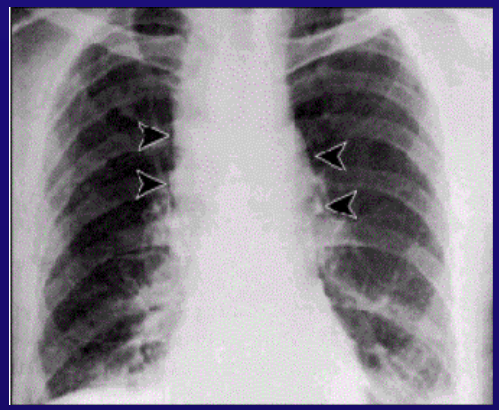
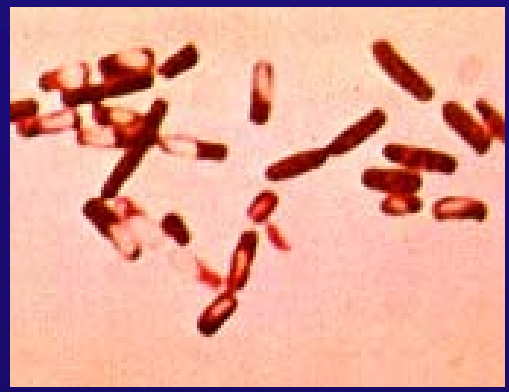
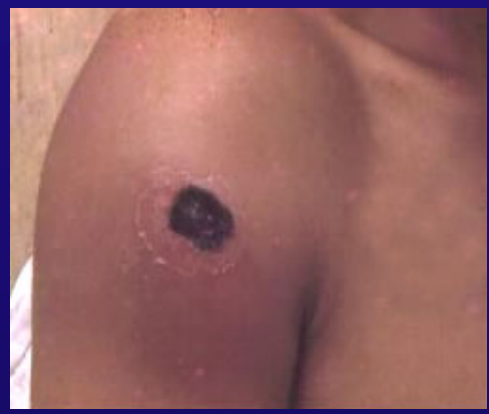
Anthrax

Bacillus anthracis







Options for post-exposure prophylaxis and therapy

- **Antibiotics (e.g., fluoroquinolone)**
- **Vaccine (e.g., AVA)**

Unmet needs (anti-toxin)

- **Antibiotics have limited efficacy in symptomatic disease**
- **Drug resistance or intolerance to antibiotics**
- **Vaccine requires time for protective immunity**

Outline

- Characterization of PA mAb
- Animal Efficacy Studies
  - Rabbit and monkey pre- and post-exposure prophylaxis
  - Rabbit post-exposure treatment
  - Rat toxin infusion model
- Phase 1 Human Study
  - Safety, pharmacokinetics and pharmacodynamics
- Next Steps
  - Additional studies
  - Manufacturing

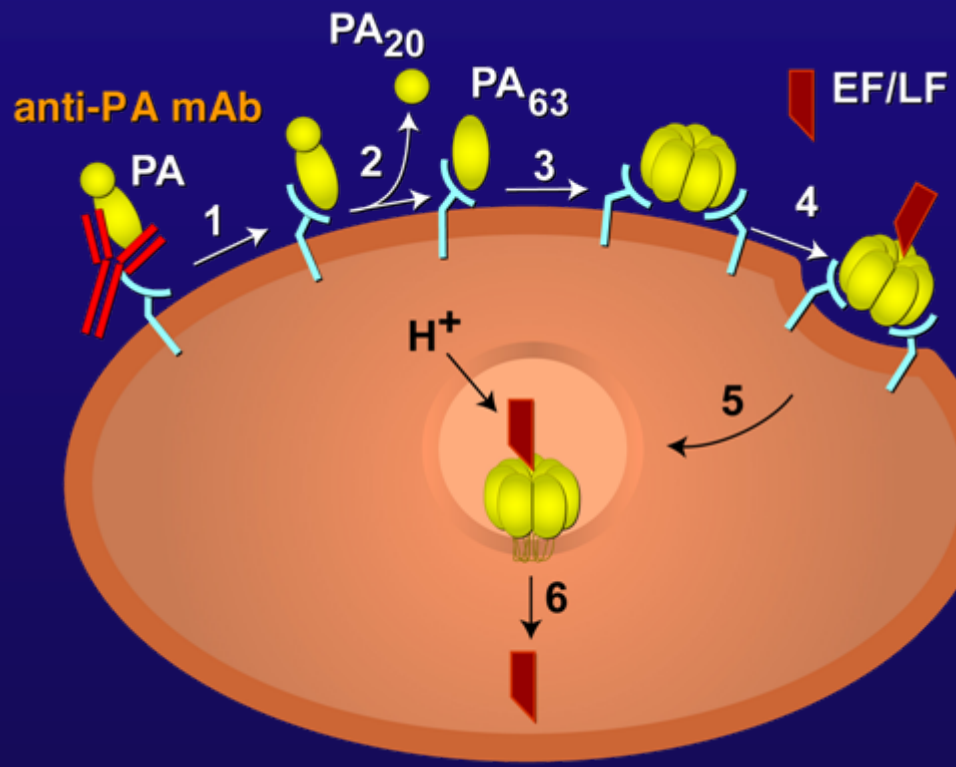
Neutralizing anti-PA Antibody

Fully human IgG1

- **Inhibits binding of PA to its receptor (high affinity and specificity)**
- **Inhibits EF mediated cAMP production (cAMP assay)**

In vivo studies

- **100% protection against toxin-mediated killing in rats**
- **Single PA mAb administration provides 100% protection for at least 3 weeks prior to toxin challenge**



modified from *Science 292:695-697, 2001*

Animal Efficacy Studies

- **Inhalational anthrax studies in rabbit and monkey**
  - **Dose that provides a survival advantage (pre-exposure or post-exposure)**
  - **Serum levels that confer protection**
  - **Improved understanding of clinical parameters reflective of disease**
- **Toxin infusion studies in rat**
  - **Hemodynamic effects of toxin**
  - **Reversal of hemodynamic effects and survival**

Rabbit and Monkey PK Supports Evaluation of a Single Injection

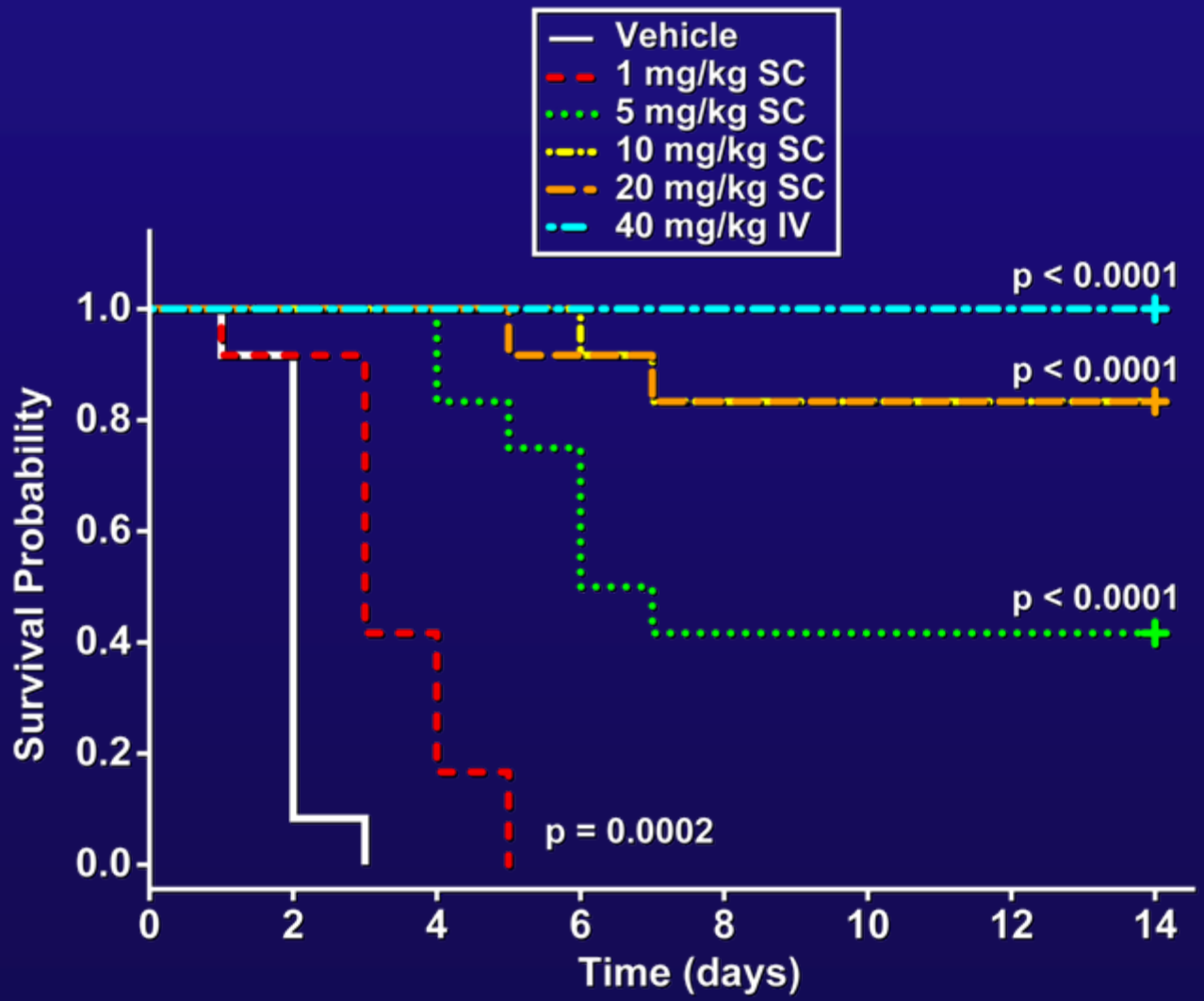
- **Cmax is similar for SC and IM routes**
  - **Rabbit (10 mg/kg) SC: 82.9 µg/mL IM: 87.8 µg/mL**
  - **Monkey (10 mg/kg) SC: 81.8 µg/mL IM: 103.7 µg/mL**
- **Good bioavailability after SC or IM injection**
- **Half life**
  - **Rabbit 7-9 days**
  - **Monkey 11-14 days**
- **Mean volume of distribution is about 78-92 mL/kg**
  - **Larger than the plasma volume (~40 mL/kg), but less than the extracellular fluid volume**
  - **Similar to other antibodies**

Study Design

- Target 100 x LD50 spores
- Survival at Day 14 (rabbit) or Day 28 (monkey)
- Time to Death
- Bacteremia and Temperature*
- Hematology, Serum Chemistry, Drug levels
- Gross Pathology

* therapeutic study in rabbit only

Survival Curves for Pre-exposure and Post-exposure Prophylaxis in Rabbits

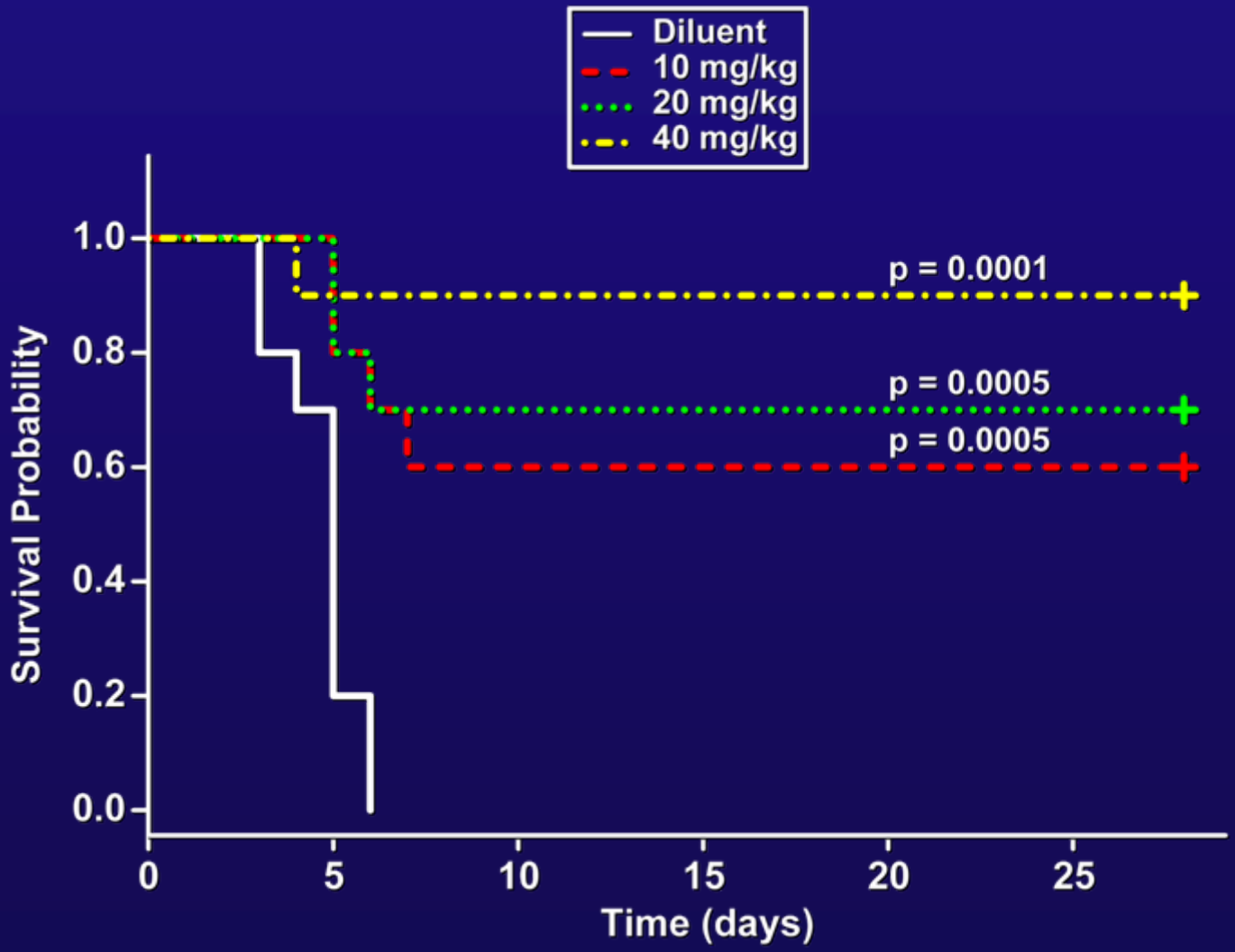


Beebe *et al*. ICAAC, 2003

Summary of Rabbit Study

- Animals were exposed to a mean spore challenge dose of 196 x LD50
- PA mAb administered SC at 10 or 20 mg/kg improves 14 day survival ($p<0.0001$)
- PA mAb administered SC prolongs time to death in a dose-dependent manner
- Incidence of bacteremia was reduced in all treatment groups
- Terminal necropsy in surviving animals showed no evidence of gross pathology (preliminary results)

Survival Curves for Pre-exposure Prophylaxis in Monkeys



Beebe *et al*. ICAAC, 2003

Summary of Monkey Study

- Animals were exposed to a mean spore challenge dose of 186 x LD50
- PA mAb administered SC at 10, 20 and 40 mg/kg improves survival ($p<0.05$)
- No bacteremia in any surviving animal at Day 7 or 14
- Surviving animals were 100% protected against inhalational spore challenge ~12 months later

Summary of PA mAb Data with Regard to Prophylaxis Spore Challenge Studies

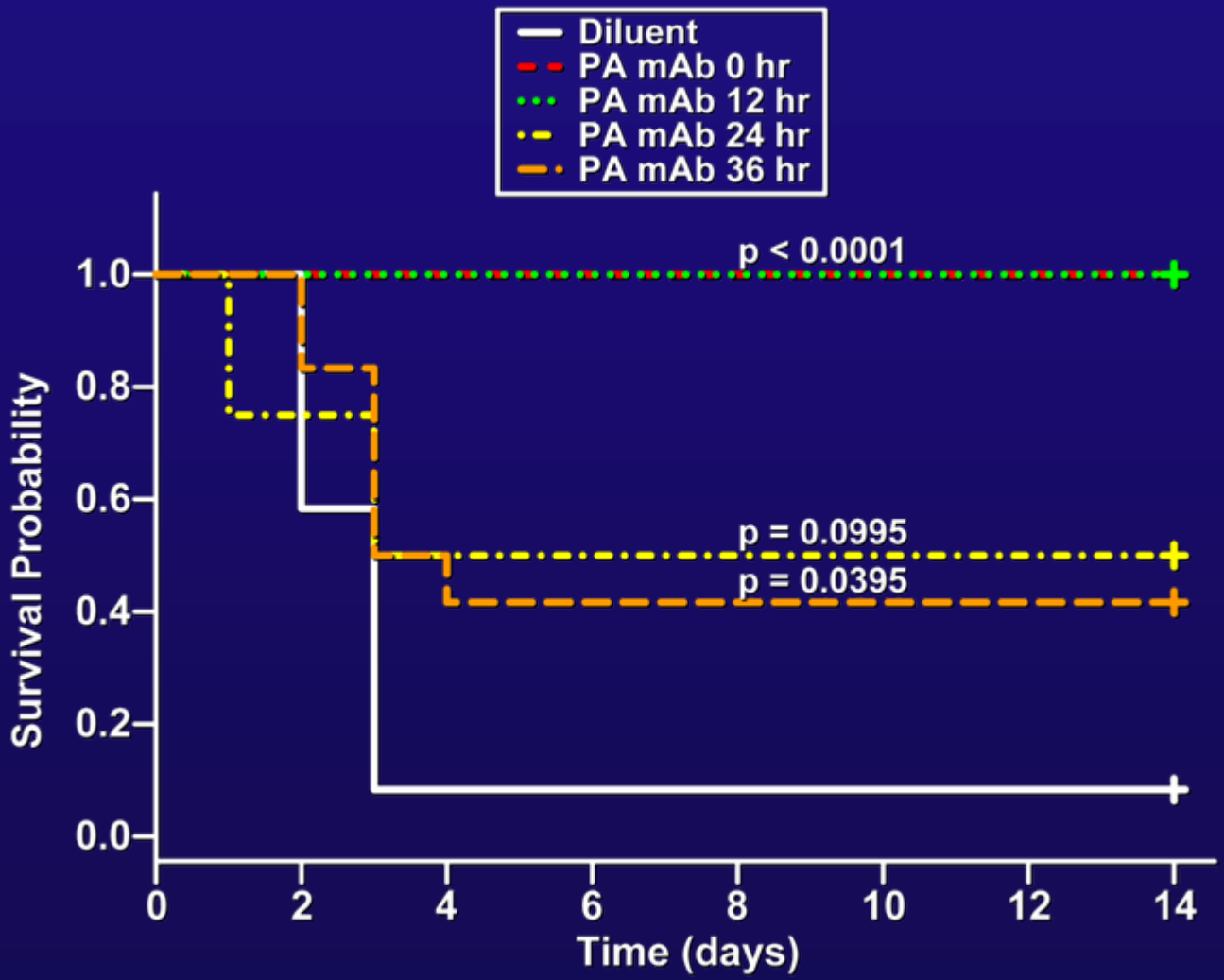
- **PA mAb (10-40 mg/kg) provided protection against anthrax spore-mediated lethality when administered to NZW rabbits 48 hrs prior to spore challenge (SC administration), or within 1 hour post-challenge (IV bolus injection)**
- **PA mAb (10-40 mg/kg) provided protection against anthrax spore-mediated lethality in cynomolgus monkeys when administered 48 hrs prior to spore challenge (SC administration)**
- **Surviving cynomolgus monkeys were protected**

Treatment Interventions

Therapeutic rescue?

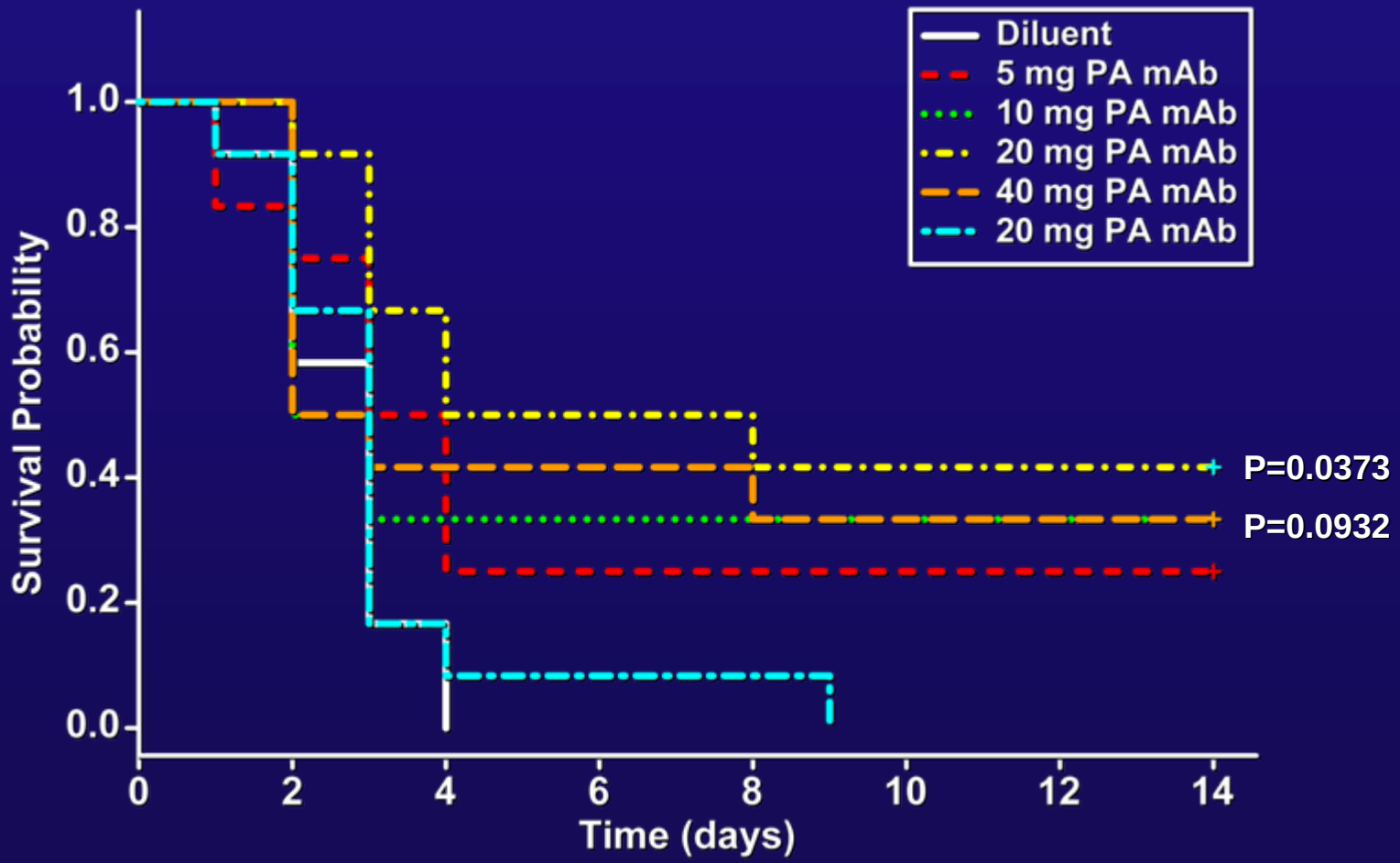
- **Challenge: What constitutes therapeutic intervention? Symptomatic disease?**
- **How does one characterize symptomatic disease or time course of disease in an animal?**
  - **Onset of bacteremia**
  - **Onset of increased temperature over baseline value**
  - **Onset of abnormal clinical observations**
- **How robust are these endpoints?**

Survival Curves for Post-exposure Treatment (0 to 36 hours) in Rabbits



Beebe *et al.* ASM, 2004

Survival Curves for Post-exposure Treatment (24 and 36 hours) in Rabbits



Beebe *et al*. ASM, 2004

Summary of Rabbit Therapeutic Studies

- PA mAb administered IV 12-24 hours post exposure at 10, 20 and 40 mg/kg improves survival
- Incidence of bacteremia increased significantly between 24 and 36 hrs post challenge
- Increase in body temperature (1F over baseline) was observed in rabbits, but was not uniformly increased in rabbits that died (vehicle treated controls)
- Time to death ranged from 1-4 days post exposure
- Because the range of “time to death” is variable, a single time point for intervention is difficult to establish

Therapeutic Rescue Studies in Rabbit: Lessons Learned

- **Onset of peripheral bacteremia gives a window of possible intervention times (24-36 hrs post challenge)**
- **Increase in body temperature is not uniformly predictive of anthrax-mediated lethality**
- **Lack of clinical signs in rabbits makes this a poor predictor of symptomatic disease**

Therapeutic Rescue Studies in Non Human Primate

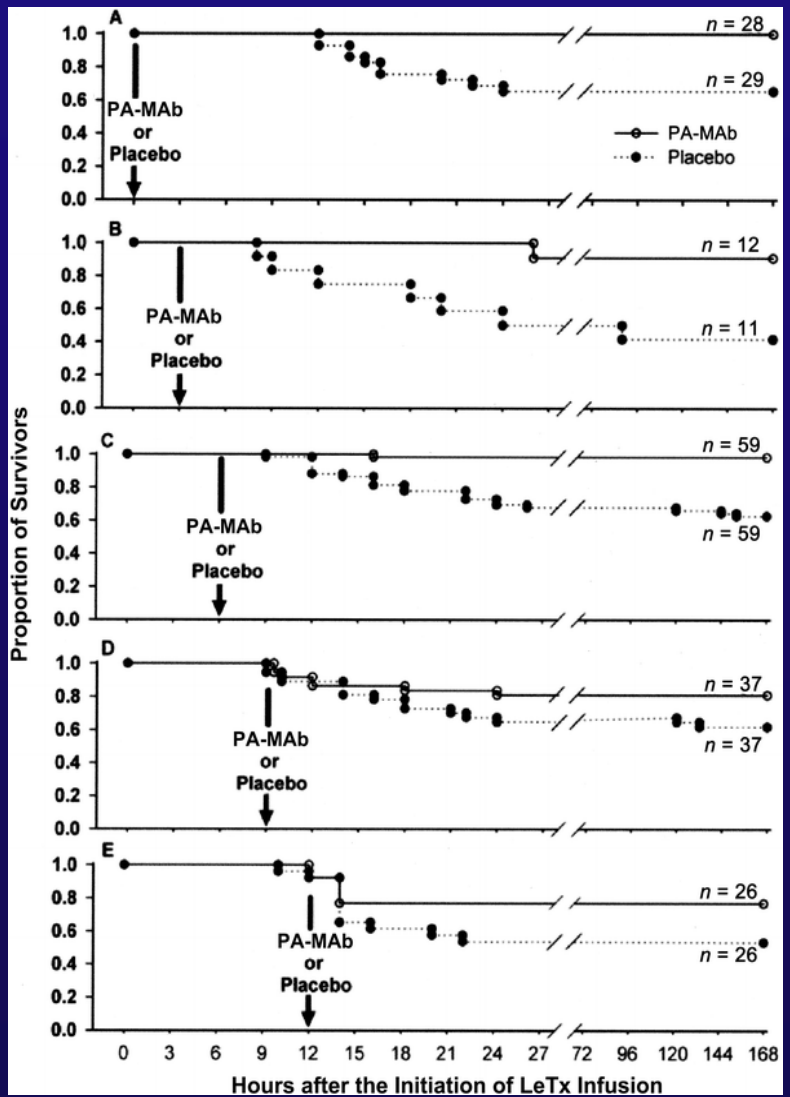
- **Lack of established therapeutic rescue model in nonhuman primates**
- **Interventions immediately after spore challenge or in the pre-symptomatic phase are considered models of post-exposure prophylaxis**
- **Evaluations of combination studies (antibiotics, vaccines, PA mAb) would likely require large number of nonhuman primates, and may not yield any more informative data than can be collected in the NZW rabbit**

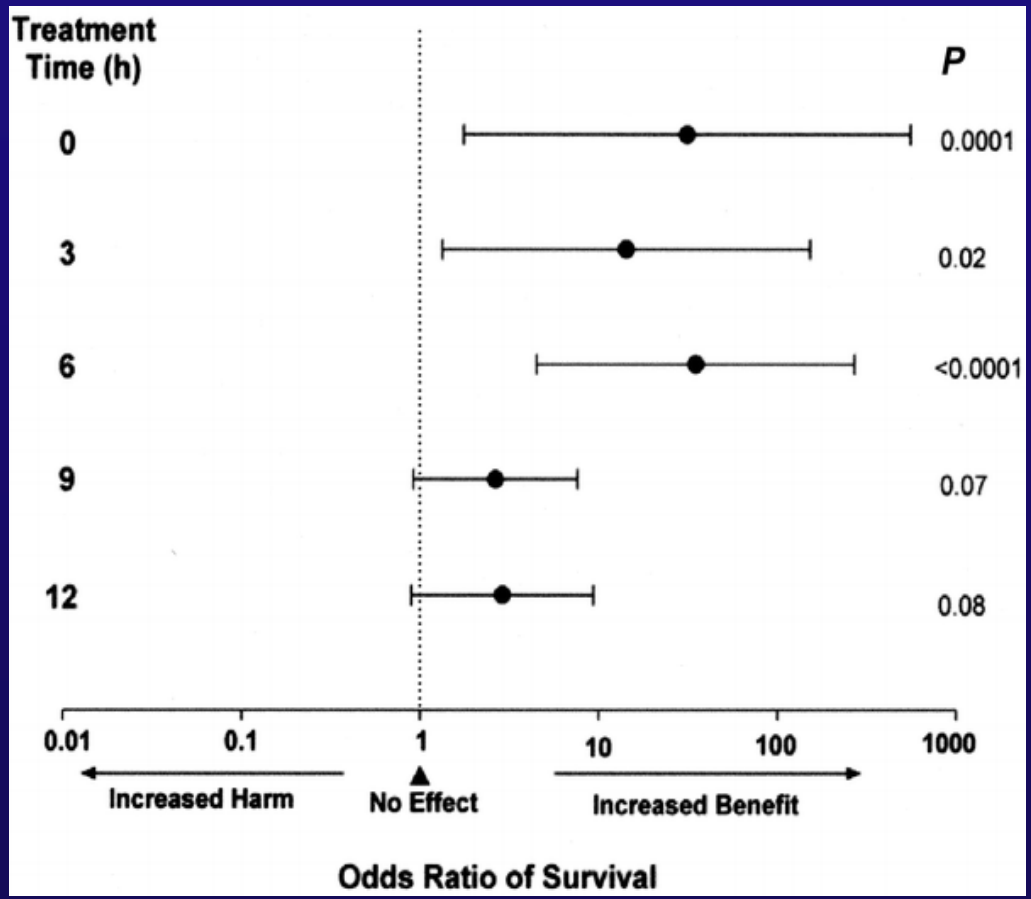
LeTx infusion model in SD rats to Evaluate Therapeutic Intervention

- **24 hr infusion allows for development of illness that may reflect accumulation of toxin after spore challenge**
- **Infusion allows for intervention times up to and including 24 hrs**
- **IV administered PA mAb demonstrated protection against LeTx lethality up to 6 hrs after initiation of infusion**
- **Hemodynamic endpoints were improved at time points 9 and 12 hrs into the infusion process**

Eichacker *et al.* JID, 2004

Survival Benefit up to 6 hours After Initiation of Toxin Infusion





Eichacker *et al.* JID, 2004

Phase 1 Study Design

Objectives:

- To evaluate the safety and pharmacokinetics of IM and IV administered PA mAb in healthy subjects
- To define the PA mAb dose for an expanded safety study in healthy subjects

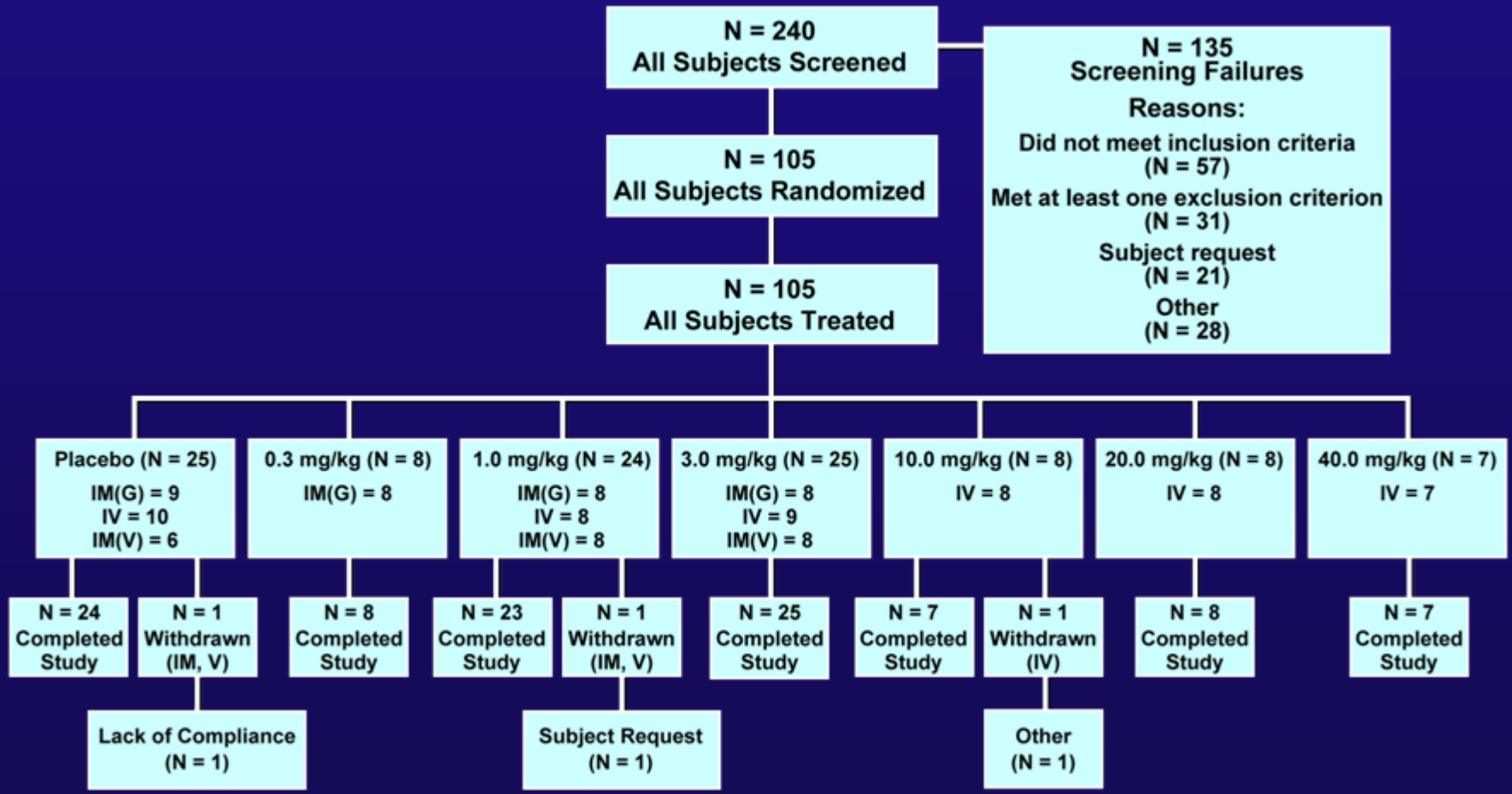
Dose Cohorts:

Single IM or IV dose of PA mAb or placebo (56 day follow-up)

IM (Gluteus or Vastus)	0.3 mg/kg, 1 mg/kg, 3 mg/kg
IV infusion	1, 3, 10, 20 and 40 mg/kg

Subramanian *et al,* CID 2005

Subject Disposition



Summary of Adverse Advents by MedDRA Preferred Term

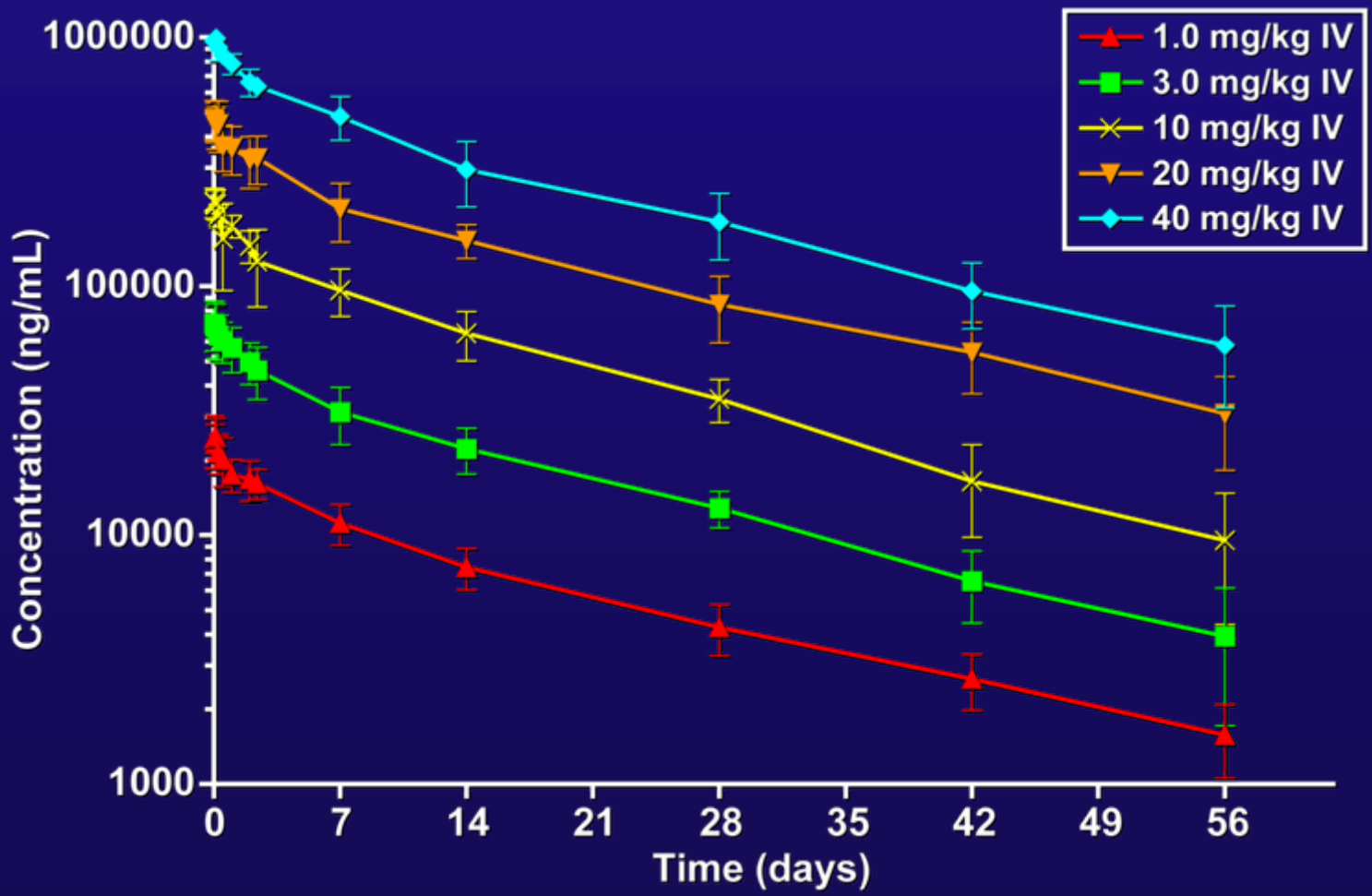
MedDRA Preferred Term	All Placebo[a] N = 25	IM Gluteus Maximus 0.3 mg/kg N = 8	IM Gluteus Maximus 1 mg/kg N = 8	IM Gluteus Maximus 3 mg/kg N = 8	IV 1 mg/kg N = 8	IV 3 mg/kg N = 9	IV 10 mg/kg N = 8	IV 20 mg/kg N = 8	IV 40 mg/kg N = 7	IM Vastus Lateralis 1 mg/kg N = 8	IM Vastus Lateralis 3 mg/kg N = 8
Headache	6 (24.0%)	1 (12.5%)	2 (25.0%)	1 (12.5%)	2 (25.0%)	3 (33.3%)	2 (25.0%)	1 (12.5%)	3 (42.9%)	-	-
Cough	1 (4.0%)	1 (12.5%)	2 (25.0%)	2 (25.0%)	-	1 (11.1%)	-	1 (12.5%)	-	-	3 (37.5%)
Nausea	-	-	-	-	1 (12.5%)	1 (11.1%)	-	-	1 (14.3%)	2 (25.0%)	-
Vomiting	-	-	-	-	1 (12.5%)	-	-	1 (12.5%)	1 (14.3%)	2 (25.0%)	-
Arthralgia	1 (4.0%)	-	-	-	2 (25.0%)	-	1 (12.5%)	-	1 (14.3%)	1 (12.5%)	-
Pyrexia	-	-	-	1 (12.5%)	-	1 (11.1%)	-	-	-	1 (12.5%)	1 (12.5%)
Nasal congestion	2 (8.0%)	-	-	1 (12.5%)	-	-	-	-	2 (28.6%)	-	1 (12.5%)
Throat irritation	1 (4.0%)	-	-	1 (12.5%)	-	1 (11.1%)	-	-	1 (14.3%)	-	1 (12.5%)
Abdominal pain	-	-	-	-	-	-	-	1 (12.5%)	1 (14.3%)	1 (12.5%)	-
Pain	-	-	-	1 (12.5%)	1 (12.5%)	-	1 (12.5%)	-	-	-	-
Upper respiratory tract infection	2 (8.0%)	-	1 (12.5%)	-	1 (12.5%)	-	-	-	-	-	1 (12.5%)
Pain in extremity	1 (4.0%)	-	-	-	-	-	2 (25.0%)	-	-	1 (12.5%)	-
Dizziness	-	-	-	-	-	1 (11.1%)	-	-	1 (14.3%)	1 (12.5%)	-
Paraesthesia	-	-	-	-	-	1 (11.1%)	2 (25.0%)	-	-	-	-
Rhinorrhoea	-	1 (12.5%)	-	1 (12.5%)	-	-	-	-	-	-	1 (12.5%)

[a]Venipuncture site pain was also reported for 2 placebo subjects (8.0%) but was not included in this table because it did not occur in ≥ 3 PA mAb subjects. All other AEs in the placebo group were reported for 1 subject each (4.0%).

Safety and Tolerability

- Safe and well tolerated with only mild to moderate adverse events reported
- No statistically significant difference in AE profiles between active and placebo or IV and IM routes of administration
- No dose or route-related increase in adverse events were observed
- No significant laboratory abnormalities were reported
- One SAE was reported – Pyelonephritis, considered not related to study drug

Mean Serum Concentrations in Subjects Administered 1 to 40 mg/kg PA mAb by 2-hour Infusion

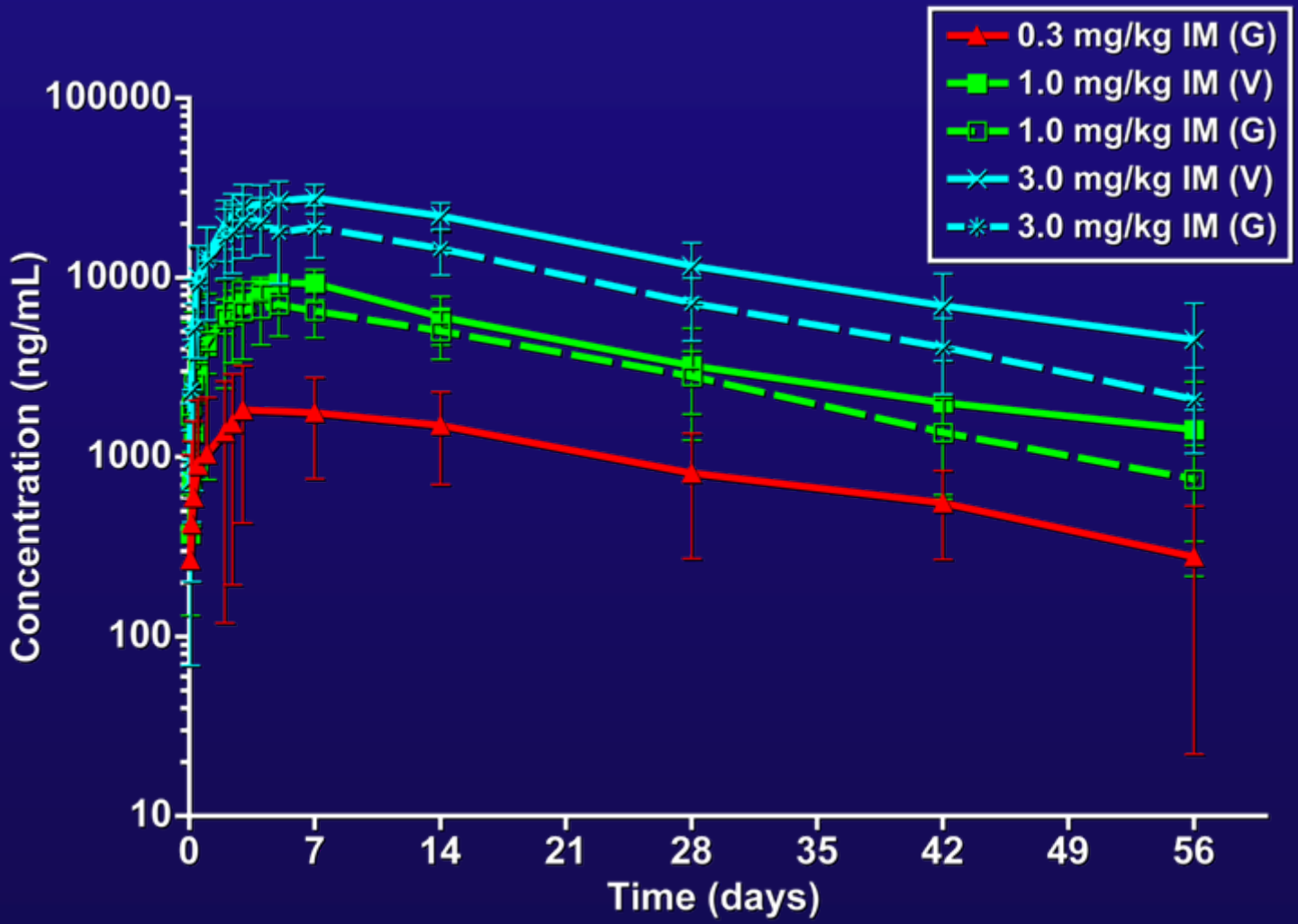


PK Parameters Following IV Dosing with 1, 3, 10, 20, or 40 mg/kg PA mAb

Dose	N = 8 Mean ± SD 1.0 mg/kg	N = 9 Mean ± SD 3.0 mg/kg	N = 8[a] Mean ± SD 10.0 mg/kg	N = 8 Mean ± SD 20.0 mg/kg	N = 7 Mean ± SD 40.0 mg/kg
$t_{½}$ (h)	464.5 ± 66.4	447.6 ± 103.1	388.0 ± 03.2	452.3 ± 78.2	389.0 ± 55.2
C_{max} (ng/mL)	25466 ± 5356	72429 ± 16909	225569 ± 24986	481078 ± 91758	1042383 ± 87509
C_{max}/dose (kg/mL)	0.0255 ± 0.0054	0.0241 ± 0.0056	0.0226 ± 0.0025	0.0241 ± 0.0046	0.0261 ± 0.0022
AUC/dose (h/mL*kg)	8.82 ± 1.79	8.59 ± 1.71	7.15 ± 1.23	8.66 ± 2.28	9.33 ± 1.96
CL (mL/kg/h)	0.12 ± 0.020	0.12 ± 0.02	0.14 ± 0.02	0.12 ± 0.03	0.11 ± 0.02

[a] C_{max} was determined for all subjects. Terminal phase parameters could not be calculated for 1 subject (053) in Cohort 6.

Mean Serum Concentrations in Subjects Administered 0.3 to 3 mg/kg PA mAb by IM Injection in the Gluteus Maximum (G) or Vastus Lateralis (V)



Mean (± SD) PK Parameters Following IV Dosing with 0.3, 1, or 3 mg/kg of PA mAb

Dose (site)	N = 8[a] Mean ± SD 0.3 mg/kg (G)	N = 8[a] Mean ± SD 1.0 mg/kg (G)	N = 8 Mean ± SD 3.0 mg/kg (G)	N = 8 [a] Mean ± SD 1.0 mg/kg (V)	N = 8 Mean ± SD 3.0 mg/kg (V)
$t_{½}$ (h)	463.3 ± 78.9	376.6 ± 100.1	362.8 ± 56.5	427.9 ± 250.3	432.8 ± 127.7
C_{max} (ng/mL)	1975 ± 1294	8022 ± 3031	22540 ± 7104	9789 ± 1600	28521 ± 6644
C_{max}/dose (kg/mL)	0.0066 ± 0.0043	0.0080 ± 0.0030	0.0075 ± 0.0023	0.0098 ± 0.0016	0.0095 ± 0.0022
AUC/dose(h/mL*kg)	6.04 ± 2.55	4.73 ± 1.59	4.27 ± 1.44	6.27 ± 3.06	7.30 ± 2.41
V_z/F (mL/kg)	128 ± 53	140 ± 111	135 ± 49	97 ± 18	87 ± 14
CL/F (mL/kg/h)	0.21 ± 0.12	0.25 ± 0.14	0.26 ± 0.10	0.21 ± 0.13	0.15 ± 0.05

[a] C_{max} and t_{max} were determined for all subjects. Terminal phase parameters could not be calculated for 1 subject in Cohort 2 and 2 subjects in each of Cohorts 1 and 9.

[b] t_{lag} and t_{max} are summarized as median and range.

G – Gluteus maximus

V – Ventrolateral thigh

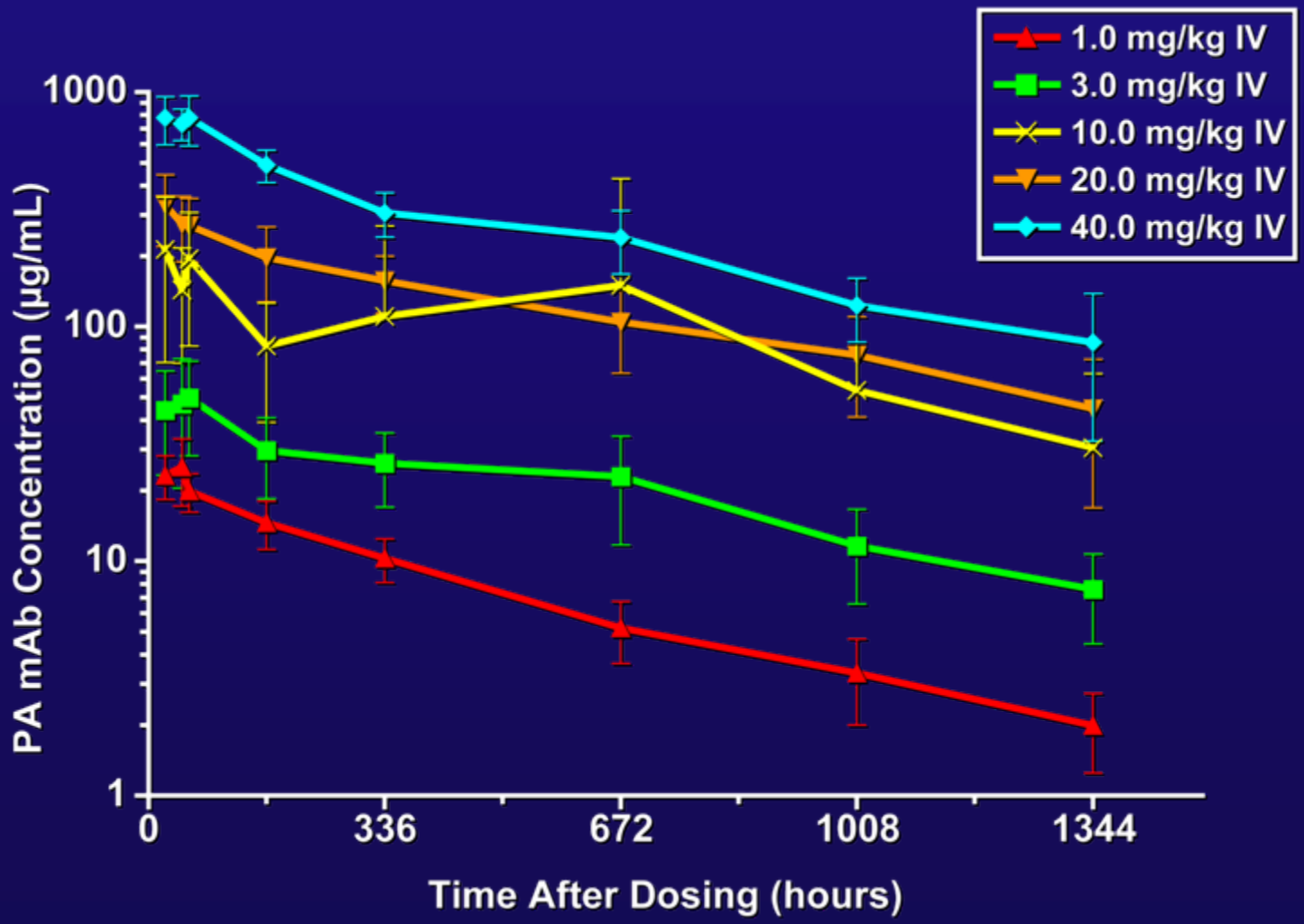
Comparison of Pharmacokinetic Parameters Across the 2 IM Injection Sites

	Mean ± SE		
	Gluteus Maximus	Vastus Lateralis	P-Value
Dose-normalized C_{max} (kg/mL)	0.0074 ± 0.0007	0.0096 ± 0.0005	0.0159
Dose-normalized $AUC_{0-\infty}$ (h*kg/mL)	4.93 ± 0.42	6.86 ± 0. 71	0.0175
CL/F (mL/h/kg)	0.24 ± 0.03	0.18 ± 0.02	0.0856
V_z/F (mL/kg)	134.6 ± 15.9	91.6 ± 4.3	0.0372

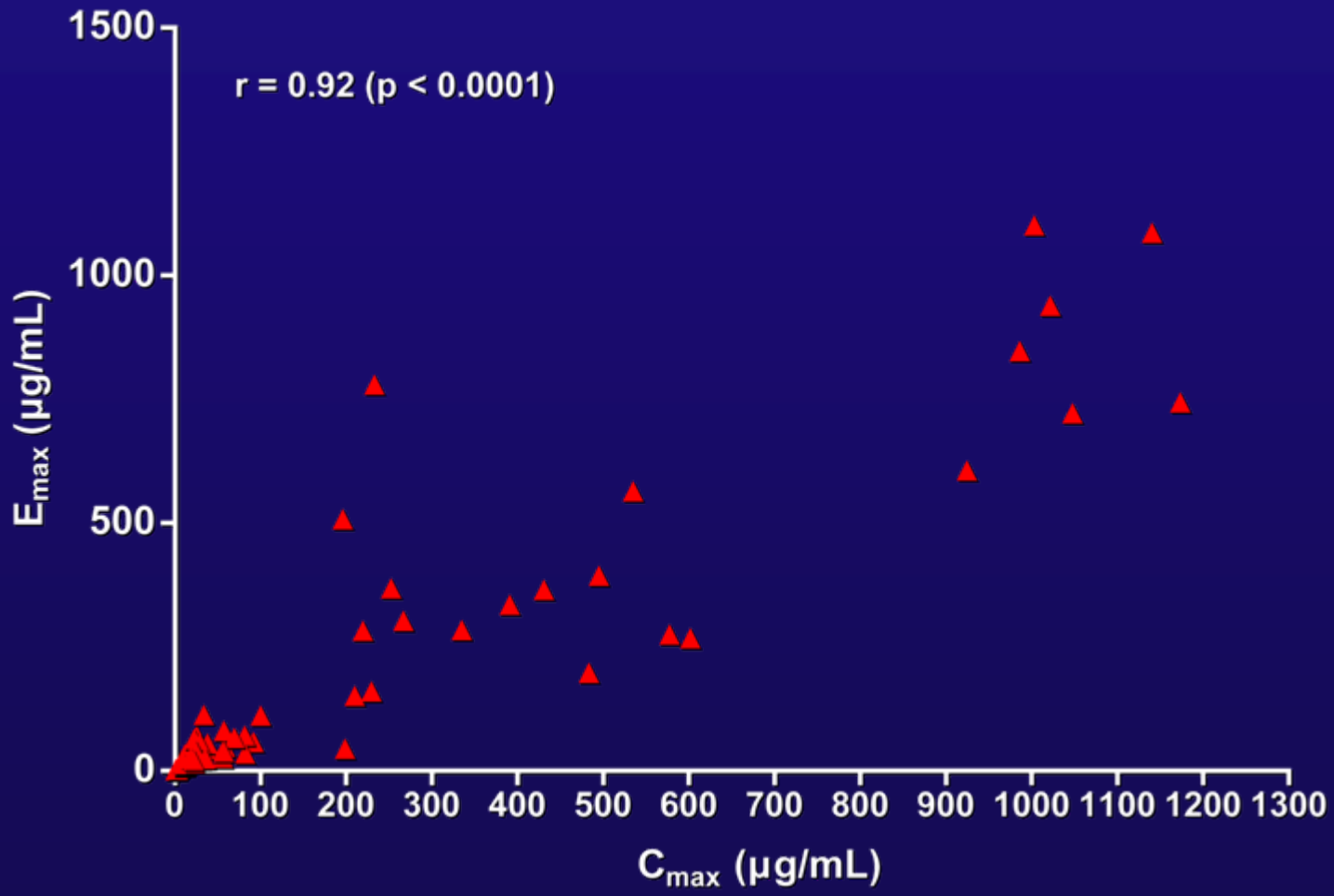
Summary: Pharmacokinetics

- **PA mAb half-life**
  - **IM: 15 to 22 days and t_{max} is between 4.5 and 7.5 days**
  - **IV: 17-21 days**
- **A greater C_{max}, AUC and bioavailability are achieved following IM injection given in the anterolateral thigh compared to the gluteus**
- **Bioavailability in the anterolateral thigh is 83% to 90%**
- **Pharmacokinetics are linear from 0.3 to 3 mg/kg after IM dosing and between 1 mg/kg and 40 mg/kg for IV dosing**

Mean Serum PA mAb Bioactivity in Subjects Dosed with IV 1 to 40 mg/kg PA mAb



E_{max} Obtained Using the Bioassay Versus C_{max} from the PK ELISA



Conclusions

1. PA mAb is a novel fully human monoclonal antibody directed against the Protective Antigen of *B. Anthracis*
2. PA mAb is safe and well tolerated when administered either IV or IM in humans
3. IV administered PA mAb exhibits a biphasic elimination profile with a prolonged half-life of 15 to 20 days in humans
4. PA mAb concentrations comparable to, or in excess of, anti-PA Ab levels that correlated with significant protection in animal models of inhalational anthrax
5. PA mAb bioactivity (as measured by cAMP bioassay) correlates well with PA mAb plasma levels
6. Based on human safety and animal efficacy data, PA mAb represents a promising candidate for the treatment and prophylaxis of inhalational anthrax

Additional Studies

- **Animal studies**
  - **PA mAb and fluoroquinolone combination study to evaluate efficacy**
- **Human studies**
  - **Expanded PA mAb safety program in a larger population**
  - **PAmAb in combination with fluoroquinolone to determine if the serum levels of either agent are impacted by the combination treatment**
  - **PA mAb in combination with vaccination (AVA) to evaluate effects of PA mAb on the development of anti-PA polyclonal response**

Manufacturing Capabilities from 1600 L to 20,000 L scale

HGS has been awarded a two-phase contract to supply PA mAb for the treatment of inhalation anthrax disease to the U.S. Government

Ten grams of PA mAb has been supplied to the U.S. Government for *in vitro* and *in vivo* testing

The U.S. Government has an option to place an order for supply of PA mAb for the Strategic National Stockpile